Figure 2. Best substrates identified by screening tyrosine library sub-sets 1 to 5 (SEQ ID NOS:4-7, respectively, in order of appearance) against ZAP-70 protein tyrosine kinase. The protein tyrosine kinase library described in Figure 1 was phosphorylated for 30 minutes at 30^{0}C using the catalytic domain of human ZAP-70. Peptides were captured using strepavidin-coated 96 well plates and phosphotyrosine detected using anti-phosphotyrosine antibody, anti mouse IgG-HRP and tetramethylbenzidine (see experimental methods). Best substrates were identified as those which gave the highest amount of phosphate incorporation.

The paragraph beginning at Page 8, line 15:

Figure 3. Km Determination of Biotin-εAHA-DEEDYFE(Nle) (SEQ ID NO:1). The catalytic domain of human ZAP-70 was used to phosphorylate varying concentrations of peptide for 10 minutes at 30^{0}C in the presence of ^{33}P-γ-ATP. Peptide capture was performed using strepavidin filter plates, scintillation fluid added, and counting performed using a beta-counter (see experimental methods). Samples were assayed in triplicate.

The paragraph beginning at Page 9, line 16:

wherein each library subset is of formula (I)

$(Xaa)_x$ Zaa $(Xaa)_y$ (I)

The paragraph beginning at Page 10, line 5:

This invention can be applied for instance to a protein tyrosine kinase in order to exemplify the technology. It provides a rapid method of identifying discrete protein kinase substrate sequences which allows pharmacophore generation and design of active site inhibitors. This invention can also be used to directly identify protein kinase inhibitor molecules. General formula: $(Xaa)_x$ Tyr $(Xaa)_y$.

The paragraph beginning at Page 11, line 25:

The results obtained from the library screen clearly demonstrated amino acid residues preferred by the protein kinase at each of the -4 to +4 sites (Figure 2). The 5 mer peptides overlapped to give information on amino acid preference at each of the binding positions -4 to +4. To confirm this a consensus peptide, Biotin-∈AHA-DEEDYFE(Nle) (SEQ ID NO: 1), representing the best -4 to +4 amino acids was made and tested as a substrate (Figure 3). This substrate gave a Km against ZAP-70 of 15.79 μM, which is better than the best ZAP-70 substrate described in the literature, a longer peptide of 14 amino acids with a tag of 3 arginines and a K_m of 29 μM (Wandenburg *et al*, 1996).

Page 13, line 18, amend to read:

General formula: $(Xaa_x\ Ser\ (Xaa)_y$.

Page 19, line 6, amend to read:

5' CCGGGATCCGCCATGCCCATGGACACGAGCGTGTAT 3' (SEQ ID NO: 2)

Page 19, line 9, amend to read:

CTTCTGTGT 3' (SEQ ID NO:3)

The paragraph beginning at Page 20, line 4:

Following homogenisation using a dounce pestle B, the cleared lysate was loaded onto a cobalt-sepharose column. After column washing with lysis buffer, elution was performed with an imidazole gradient and ZAP-70 fractions identified by protein kinase activity against the peptide substrate (SEQ ID NO:19) Lys-Lys-Lys-Lys-Ala-Asp-Glu-Glu-Asp-Tyr-Phe-Ile-Pro-Pro-Ala as described in Casnellie *et al*, 1991.

The paragraph beginning at page 20, line 28:

The best substrates were identified as those which gave the highest amount of phosphate incorporation. The library subsets were deconvoluted according to the teaching of W097/42216: this gives an immediate determination of the

unique sequence of any phosphorylated motif without the need for further synthesis or sequencing. (Figure 2 (SEQ ID NOS:4-7, respectively, in order of appearance).

Page 21, line 25, amend to read:

Library Sub-Set 1 Asp-Glu-Glu-Asp-Tyr (SEQ ID NO:8)

Page 21, line 26, amend to read:

Library Sub-Set 2 Asp-Glu-Glu-Tyr-Asp (SEQ ID NO:9)

Page 22, line 1 amend to read:

Library Sub-Set 3 Asp-Glu-Tyr-Glu-Asp (SEQ ID NO:10)

Page 22, line 2 amend to read:

Library Sub-Set 4 Asp-Tyr-Glu-Glu-Val (SEQ ID NO:11)

Page 22, line 3 amend to read:

Library Sub-Set 5 Tyr-ser-Ile-Ile-Nle (SEQ ID NO:12)

Page 22, line 12 amend to read:

Library Sub-Set 1 Asp-Glu-Glu-Glu-Tyr (SEQ ID NO:13)

Page 22, line 13 amend to read:

Library Sub-Set 2 Asp-Glu-Glu-Tyr-Phe (SEQ ID NO:14)

Page 22, line 14 amend to read:

Library Sub-Set 3 Asp-Glu-Tyr-His-Asn (SEQ ID NO:15)

Page 22, line 15 amend to read:

Library Sub-Set 4 Asp-Tyr-His-Leu-Phe (SEQ ID NO:16)

Page 22, line 16 amend to read:

Library Sub-Set 5 Tyr-Pro-Ile-Glu-Val (SEQ ID NO:17)

The paragraph beginning at Page 22, line 22:

In this example the invention was used to map the active catalytic site of v-Abl, a protein kinase enzyme that catalyses the phosphorylation of a tyrosine residue. For this enzyme only the Library Sub-Set 4 (i.e. X-Tyr-X-X-X) was scanned with the enzyme. The active site substrate recognition substrate for this enzyme for this Sub-Set was Serine-tyrosine-phenylalanine-histamine-glutamine (SEQ ID NO:18).

Page 26, please insert the attached sequence listing pages and renumber